transactivation domain is dependent on protein phosphorylation and/or protein:protein interaction, and wherein binding of said fusion protein to said recognition sequence results in transactivation of said reporter gene when said transactivation domain fused to said DNA binding domain is activated.

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38. ~~32.~~ (New) The kit of claim 25, said kit further comprising a nucleic acid expression construct encoding an upstream activator of the conditionally active transactivation domain.

REMARKS

Claims 1-9 and 24-32 are currently pending in the application. Claim 25 is amended. New claims 27-32 are added. The amendments and new claims are made to correct claims 25-26, which depended from non-elected claims 10, 11, 17 and 18. Claim 25, which depended from non-elected claims 10, 11, 17 and 18, is amended herewith to independent form and now incorporates the required material from claim 10. New claims 27, 29 and 31 are also written in independent form, and represent claim 25, with the inclusion of the subject matter of claims 11, 17 and 18, respectively. The amendments and new claims therefore find support in the specification and the claims as originally filed. No new matter is added.

Claim Rejections Under 35 U.S.C. § 103

Claims 1-9 and 24-26 are rejected under 35 U.S.C. § 103(a) as being unpatentable over Montminy (U.S. Pat. No. 6,063,583) in view of Gilman *et al.* (U.S. Pat. No. 6,306,649).

Montminy teaches the use of three constructs to identify compounds which disrupt the formation of a complex between CREB and CBP. The three constructs are: (1) a reporter construct which includes a GAL4 response element operatively linked to a reporter gene, (2) a fusion protein which contains a GAL4 DNA binding protein operatively linked to the Kinase Inducible Domain (KID) of CREB, and (3) a fusion protein containing an activation domain operatively linked with the KIX binding domain of CBP. When the KIX domain of construct (3)

binds to the phosphorylated CREB of construct (2), the GAL4 DNA binding protein then binds to the GAL4 response element of construct (1), which then induces the reporter gene. When a compound is added which disrupts the complex of CREB and CBP, the reporter gene is not activated.

In contrast, the present invention discloses cell lines and kits, which use: (1) a reporter gene operably linked to a recognition sequence for a sequence-specific DNA-binding protein, and (2) a fusion protein containing a sequence-specific DNA binding domain, and a conditionally active transactivation domain of CREB. The sequence-specific DNA binding domain specifically binds to the recognition sequence, and the binding results in transactivation of the reporter gene.

Montminy teaches the use of three constructs, while the present claims recite two. In addition, Montminy teaches that the GAL4 response element activates the reporter gene, while the present claims recite that the transactivation domain of CREB activates the reporter gene. Therefore, Montminy teaches upstream, or cis-activation, while the present claims specifically recite transactivation from the other construct.

The Office Action states that Montimy does not specifically teach stable integration of the constructs into the cell, and relies upon Gilman *et al.* for this aspect. However, the combination of the two references simply teaches the stable integration into a cell of the three constructs of Montminy, and therefore still produces a three-construct method of cis-activation of a reporter gene, in direct contrast with the teaching of the present claims of a two-construct method of transactivation of a reporter gene. Use of other reporter genes, binding domains, expression systems, and cell lines do not remedy the differences between the present invention and the combination of the teachings of the cited references.

Applicant therefore respectfully submits that the claims are not obvious in view of the cited references, and respectfully requests that the rejection on this basis be reconsidered and withdrawn.

Double Patenting

Claims 1-9 and 24-26 are provisionally rejected under the judicially-created doctrine of obvousness-type double patenting as being unpatentable over claims 1-9 and 24-26 of copending U.S. App. No. 09/637,511.

Upon the indication of allowable subject matter, Applicant will provide a Terminal Disclaimer limiting the term of any patent issuing from the present applcation to that of the patent issuing from U.S. App. No. 09/637,511.

Applicant submits that in view of the foregoing remarks, all issues relevant to patentability raised in the Office Action have been addressed. Applicant respectfully requests the withdrawal of rejections over the claims of the present invention.

Respectfully submitted,

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MARKED-UP VERSION OF AMENDMENTS:

Specification Amendments Under 37 C.F.R. § 1.121(b)(1)(iii)

Please replace the paragraph at page 15, lines 11 through 19 with the paragraph below, which is marked up by way of bracketing and underlining to show the changes relative to the previous version of the paragraph:

As used herein, the term "sequence-specific DNA binding protein" refers to a protein that recognizes and binds a specific DNA sequence. The sequence bound by a sequence-specific DNA binding protein may be an invariant arrangement of contiguous nucleotide residues (e.g., GGATCC; SEQ ID NO:1) or it may be a conserved sequence motif in which individual residues may vary and still allow recognition and binding by the sequence-specific DNA binding protein (e.g., GGPuPyCC (SEQ ID NO:2), wherein Pu and Py are purine and pyrimidine, respectively). Binding of the protein to its specific sequence may be assessed via any conventional protein:nucleic acid binding methods, including but not limited to electrophoretic gel analysis of a given protein:nucleic acid construct.

Please replace the paragraph at page 20, lines 4 through 7 with the paragraph below, which is marked up by way of bracketing and underlining to show the changes relative to the previous version of the paragraph:

A schematic diagram of the pFR-Luc reporter vector is shown. Five copies of the GAL4 DNA-binding domain recognition sequence (underlined; SEQ ID NOs:3, 4, 5, 6, 7) are linked to a minimal promotor containing a TATA element upstream of the initiator ATG initiator codon of firefly luciferase coding sequences.

Please replace the paragraph at page 20, lines 10 through 14 with the paragraph below, which is marked up by way of bracketing and underlining to show the changes relative to the previous version of the paragraph:

A schematic diagram of the plasmid pFA-CMV, used as the base vector for the fusion transactivator plasmids according ro the invention is shown. The vector fuses the GAL4 DNA-binding domain (amino acids 1-147) to the selected fusion transactivation domain via the shown multiple cloning site (SEQ ID NOs:8, 9). Expression of the resulting fusion transactivator protein is driven by the strong CMV promoter.

Please replace the paragraph at page 36, lines 1-7 with the paragraph below, which is marked up by way of bracketing and underlining to show the changes relative to the previous version of the paragraph:

To facilitate the integration and selection for stable reporter gene integration, a hygromycin resistance expression cassette, excised from p3'SS (a vector for LacSwitch™ expression systems (Stratagene), GenBank Accession No. U42371), was inserted into the Ndel site of the pFR-Luc (Genbank Accession No. AF058756) luciferase reporter vector, to generate pFR-Luc-Hyg. pFR-Luc (and therefore pFR-Luc-Hyg) carries five copies of the GAL4 DNA-binding domain recognition sequence 5'-CGGAGTACTGTCCTCCG-3' (SEQ ID NO:10) upstream of a basic TATA element and the coding region for firefly luciferase (see Figure 4).

Claim Amendments Under 37 C.F.R. § 1.121(c)(1)(ii)

Please amend claim 25 as follows:

25. (Amended) A kit for performing a [the] method of [any one of claims 10, 11, 17 or 18,] assaying for the activity of a signal transduction pathway in a mammalian cell, said method comprising the steps of:

detecting in a signal transduction pathway-specific reporter cell line expression of a reporter gene, wherein said reporter cell line comprises:

a reporter gene operably linked to a recognition sequence for a sequence-specific DNA-binding protein; and

a stably integrated recombinant nucleic acid construct comprising a sequence encoding a fusion protein, said fusion protein comprising:

a sequence-specific DNA binding domain, wherein said DNA binding domain specifically binds said recognition sequence; and

a conditionally active transactivation domain;

wherein activation of said conditionally active transactivation domain is dependent on protein phosphorylation and/or protein:protein interaction, wherein binding of said fusion protein to said recognition sequence results in transactivation of said reporter gene when said transactivation domain fused to said DNA binding domain is activated, wherein expression of said reporter gene is indicative of activity of said signal transduction pathway;

said kit comprising a cell line comprising a stably integrated recombinant nucleic acid construct comprising:

a reporter gene operably linked to a recognition sequence for a sequence-specific DNA-binding protein; and

a stably integrated recombinant nucleic acid construct comprising a sequence encoding a fusion protein, said fusion protein comprising a sequence-specific DNA binding domain, wherein said DNA binding domain specifically binds said recognition sequence, and a conditionally active transactivation domain, wherein activation of said conditionally active transactivation domain is dependent on protein phosphorylation and/or protein:protein interaction, and wherein binding of said fusion protein to said recognition sequence results in transactivation of said reporter gene when said transactivation domain fused' to said DNA binding domain is activated.